

Guidelines for Survival Rodent Surgery

Scope: These guidelines apply to all surgical procedures performed on rodents at the NIH in which the animals are expected to recover from anesthesia.¹ Prior to performing any survival surgery techniques on rodents, an approved Animal Study Proposal must be in place with appropriately trained personnel and procedures available. Specific procedures to accomplish these guidelines can be obtained from your veterinarian.

General: The following principles described in the *Guide for the Care and Use of Laboratory Animals* apply to rodent surgery.

- Appropriate pre-operative and post-operative care of animals in accordance with established veterinary medical and nursing practices are required.
- A dedicated surgical facility is not required.
- All survival surgery will be performed by using aseptic procedures, including masks, sterile gloves, sterile instruments, and aseptic techniques

The *Guide* states that it is important for research personnel to be appropriately qualified and trained in all procedures to ensure that good surgical technique is practiced.

Good technique includes:

- Asepsis,
- Gentle tissue handling,
- Minimal dissection of tissue,
- Appropriate use of instruments,
- Effective hemostasis, and
- Correct use of suture materials and *patterns or other wound closure techniques*.

Investigators should work closely with their veterinarian to assure that the challenges of multiple surgeries, for example those that occur in the production of genetically engineered mice, are adequately addressed.

Procedures:

A. Personal Protective Equipment:

1. Clean jumpsuit or lab coat
2. Mask²
3. Gloves
 - a) Sterile surgical gloves. Using sterile surgical gloves allows you to touch all areas of the sterile surgical field and surgical instruments with your gloved hand. This technique should be used whenever possible.
 - b) Clean exam gloves. Using clean exam gloves and a “tips-only” technique restricts you to using only the sterile working ends of the surgical instruments to manipulate the surgical field. The gloved, but not sterile, hand must never touch the working end of the instruments, the suture, suture needle, or any part of the surgical field. This technique is useful when working alone and manipulation of non-sterile objects (e.g., anesthesia machines, microscopes, lighting) is required (see Brown, PA & Hoogstraten, MS).
4. Hair cover

¹ A compact disk with depictions and expanded explanations of the methods recommended in these guidelines is available by sending a request to rodentcd@od.nih.gov.

² Because of the necessity of mouth pipetting, masks are not worn during embryo transfer surgeries.

B. Pre-Operative:

1. Surgery should be conducted in a disinfected, uncluttered area that promotes asepsis during surgery (see Table 1).
2. Prepare the animal by removing hair from the surgical site. Perform this procedure in an area separate from where the surgery is to be conducted.
3. Prepare the surgical site(s) with an appropriate skin disinfectant (see Table 2).
4. Surgeons should wash and dry their hands before aseptically donning sterile surgical gloves.

C. Operative:

1. The animal must be maintained in a surgical plane of anesthesia throughout the procedure.
2. Begin surgery with sterile instruments and handle instruments aseptically (see Table 3).
3. When using “tips-only” technique, the sterility of the instrument tips must be maintained throughout the procedure.
4. Instruments and gloves may be used for a series of similar surgeries provided they are maintained clean and disinfected between animals (see Table 4).
5. Monitor and maintain the animal's vital signs.
6. Close surgical wounds using appropriate techniques and materials (see Table 5).

D. Post-Operative:

1. Move the animal to a warm, dry area and monitor it during recovery. Return the animal to its routine housing only after it has recovered from anesthesia (e.g., the animal can maintain itself in sternal recumbency).
2. Provide analgesics as appropriate and approved in your Animal Study Proposal.
3. Generally, remove skin closures 10 to 14 days post-operatively.
4. Maintain a surgical record (e.g., annotate cage card with procedure and date; recording the body weight on the day of surgery is strongly recommended).

References:

- American College of Laboratory Animal Medicine Position on Rodent Surgery.
[\[http://www.aclam.org/education/guidelines/position_rodentsurgery.html\]](http://www.aclam.org/education/guidelines/position_rodentsurgery.html)
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- Bradfield, JF, Schachtman, TR, McLaughlin, RM, and Steffen, EK. 1992. Behavioral and physiological effects of inapparent wound infection in rats. *Lab Anim Sci* 42(6): 572-578.
- Brown, MJ, Pearson, PT, and Tomson, FN. 1993. Guidelines for animal surgery in research and teaching. *Am J Vet Res.* 54(9): 1544-1559.
- Brown PA and Hoogstraten-Miller S. Principles of Aseptic Rodent Survival Surgery: Parts I & 2 In: Reuter J.D. and Suckow M.A. (Eds.), *Laboratory Animal Medicine and Management*. Ithaca: International Veterinary Information Service (www.ivis.org), 2004; Document No. B2514.0604.
[\[http://www.ivis.org/advances/Reuter/brown1/chapter_frm.asp?LA=1\]](http://www.ivis.org/advances/Reuter/brown1/chapter_frm.asp?LA=1) and
[\[http://www.ivis.org/advances/Reuter/brown2/chapter_frm.asp?LA=1\]](http://www.ivis.org/advances/Reuter/brown2/chapter_frm.asp?LA=1).
- Guideline for Hand Hygiene in Health Care Settings. *Morbidity and Mortality Weekly Report*, October 25, 2002 / 51(RR16); 1-44.
- Institute of Laboratory Animal Resources, National Research Council. *Guide for the Care and Use of Laboratory Animals*. Washington, DC: National Academy Press 1996; pp 556-70.
[\[http://www.nap.edu/readingroom/books/labrats/\]](http://www.nap.edu/readingroom/books/labrats/)
- Rutala, W.A. 1996. APIC guideline for selection and use of disinfectants. *Am J Infect Control.* 24:313-42.

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Appendix

Guidelines for Survival Rodent Surgery

This appendix includes definitions, tables of information, and references as a resource for investigators.

DEFINITIONS:

ASEPTIC SURGICAL PROCEDURES: Surgery performed using procedures that limit microbial contamination so that significant infection or suppuration does not occur.

MAJOR SURGERY: Any surgical intervention that penetrates and exposes a body cavity; any procedure that has the potential for producing permanent physical or physiological impairment; and/or any procedure associated with orthopedics or extensive tissue dissection or transection.

MINOR SURGERY: Any surgical intervention that neither penetrates and exposes a body cavity nor produces permanent impairment of physical or physiologic function. Examples are superficial vascular cut down, and percutaneous biopsy.

STERILIZATION: The process whereby all viable microorganisms are eliminated or destroyed. The criterion of sterilization is the failure of organisms to grow if a growth supporting medium is supplied.

DISINFECTION: The chemical or physical process that involves the destruction of pathogenic organisms. All disinfectants are effective against vegetative forms of organisms, but not necessarily spores.

Table 1. RECOMMENDED HARD SURFACE DISINFECTANTS (e.g., table tops, equipment)

Always follow manufacturer's instructions for dilution and expiration periods.

AGENT	EXAMPLES *	COMMENTS
Alcohols	70% ethyl alcohol 85% isopropyl alcohol	Contact time required is 15 minutes. Contaminated surfaces take longer to disinfect. Remove gross contamination before using. Inexpensive.
Quaternary Ammonium	Roccal®, Quatricide®	Rapidly inactivated by organic matter. Compounds may support growth of gram negative bacteria.
Chlorine	Sodium hypochlorite (Clorox® 10% solution) Chlorine dioxide (Clidox®, Alcide®, MB-10®)	Corrosive. Presence of organic matter reduces activity. Chlorine dioxide must be fresh; kills vegetative organisms within 3 minutes of contact.
Glutaraldehydes	Glutaraldehydes (Cidex®, Cetylclide®, Cide Wipes®)	Rapidly disinfects surfaces.
Phenolics	Lysol®, TBQ®	Less affected by organic material than other disinfectants.
Chlorhexidine	Nolvasan®, Hibiclens®	Presence of blood does not interfere with activity. Rapidly bactericidal and persistent. Effective against many viruses.

* The use of common brand names as examples does not indicate a product endorsement.

Table 2. SKIN DISINFECTANTS

Alternating disinfectants is more effective than using a single agent. For example, an iodophor scrub can be alternated three times with 70% alcohol, followed by a final soaking with a disinfectant solution. Alcohol, by itself, is not an adequate skin disinfectant. The evaporation of alcohol can induce hypothermia in small animals.

AGENT	EXAMPLES *	COMMENTS
Iodophors	Betadine®, Prepodyne®, Wescodyne®	Reduced activity in presence of organic matter. Wide range of microbicidal action. Works best in pH 6-7.
Chlorhexidine	Nolvasan®, Hibiclens®	Presence of blood does not interfere with activity. Rapidly bactericidal and persistent. Effective against many viruses. Excellent for use on skin.

* The use of common brand names as examples does not indicate a product endorsement.

Table 3. RECOMMENDED INSTRUMENT STERILANTS

Always follow manufacturer's instructions for dilution, exposure times and expiration periods.

AGENT	EXAMPLES *	COMMENTS
Steam sterilization (moist heat)	Autoclave	Effectiveness dependent upon temperature, pressure and time (e.g., 121°C for 15 min. vs 131°C for 3 min).
Dry Heat	Hot Bead Sterilizer Dry Chamber	Fast. Instruments must be cooled before contacting tissue. <i>Only tips of instruments are sterilized with hot beads.</i>
Gas sterilization	Ethylene Oxide	Requires 30% or greater relative humidity for effectiveness against spores. Gas is irritating to tissue; all materials require safe airing time.
Chlorine	Chlorine Dioxide	Corrosive to instruments. Instruments must be rinsed with sterile saline or sterile water before use.
Glutaraldehydes	Glutaraldehyde (Cidex®, Cetylcode®, Metricide®)	Several hours required for sterilization. Corrosive and irritating. Instruments must be rinsed with sterile saline or sterile water before use.
Hydrogen peroxide-acetic acid	Actril®, Spor-Klenz®	Several hours required for sterilization. Corrosive and irritating. Instruments must be rinsed with sterile saline or sterile water before use.

* The use of common brand names as examples does not indicate a product endorsement.

Table 4. RECOMMENDED INSTRUMENT DISINFECTANTS

Always follow manufacturer's instructions for dilution, exposure times and expiration periods.

AGENT	EXAMPLES *	COMMENTS
Alcohols	70% ethyl alcohol 85% isopropyl alcohol	Contact time required is 15 minutes. Contaminated surfaces take longer to disinfect. Remove gross contamination before using. Inexpensive.
Chlorine	Sodium hypochlorite (Clorox® 10% solution) Chlorine dioxide (Clidox®, Alcide®)	Corrosive. Presence of organic matter reduces activity. Chlorine dioxide must be fresh. Kills vegetative organisms within 3 min. Corrosive to instruments. Instruments must be rinsed with sterile saline or sterile water before use.
Chlorhexidine	Nolvasan®, Hibiclens®	Presence of blood does not interfere with activity. Rapidly bactericidal and persistent. Effective against many viruses. Instruments must be rinsed with sterile saline or sterile water before use.

* The use of common brand names as examples does not indicate a product endorsement.

Table 5. WOUND CLOSURE SELECTION

MATERIAL *	CHARACTERISTICS AND FREQUENT USES
Polyglactin 910 (Vicryl®), Polyglycolic acid (Dexon®)	Absorbable; 60-90 days. Ligate or suture tissues where an absorbable suture is desirable.
Polydioxanone (PDS®) or, Polyglyconate (Maxon®)	Absorbable; 6 months. Ligate or suture tissues especially where an absorbable suture and extended wound support is desirable
Polypropylene (Prolene®)	Nonabsorbable. Inert.
Nylon (Ethilon®)	Nonabsorbable. Inert. General closure.
Silk	Nonabsorbable. (Caution: Tissue reactive and may wick microorganisms into the wound, so <i>silk is not recommended for skin closure</i>). Excellent handling. Preferred for cardiovascular procedures.
Chromic Gut	Absorbable. Versatile material.
Stainless Steel Wound Clips, Staples	Nonabsorbable. Requires instrument for removal.
Cyanoacrylate (Vetbond®, Nexaband®)	Skin glue. For non-tension bearing wounds.

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Suture gauge selection: Use the smallest gauge suture material that will perform adequately.

Cutting and reverse cutting needles: Provide edges that will cut through dense, difficult to penetrate tissue, such as skin.

Non-cutting, taper point or round needles: Have no edges to cut through tissue; used primarily for suturing easily torn tissues such as peritoneum or intestine.